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Lipid composition of the euphausiids *Euphausia vallentini* and *Thysanoessa macrura* during summer in the Southern Indian Ocean

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Abstract: Two species of euphausiids (*Thysanoessa macrura* Sars and *Euphausia vallentini* Stebbing) from the Southern Indian Ocean were analysed for their lipid content, fatty acid and fatty alcohol composition, in relation to developmental stage (*T.m*) and sex (*E.v*). Lipid composition showed wax esters and triacylglycerols as main neutral lipids for *T. macrura* and *E. vallentini* respectively. Allometric relationships between lipid class and total lipids indicated that changes in total lipids were size dependent and mainly related to wax esters in *T. macrura* and polar lipids (both phosphatidylcholine or PC and phosphatidylethanolamine or PE) in *E. vallentini*. No difference in lipid composition could be shown for male and female *E. vallentini*, while sampling location, developmental stage and sex were significantly influential in *T. macrura*. In this latter species wax esters displayed relatively similar fatty acid and alcohol composition in both juvenile and female stages, whereas a striking difference could be seen between females and juveniles in terms of polar lipids with a very low PE content in females. Male and female *E. vallentini* showed little differences in fatty acid structure with the exception of PC in females, which were low in 22:6n-3. The trophic status of these two species was established using multivariate discriminant analysis, which indicated for *E. vallentini* a degree of omnivory similar to *E. superba*, while these *T. macrura* appeared more omnivorous than individuals collected in other areas of the Southern Ocean. Clustering of polar lipid composition suggested a link between differences in PC fatty acid and the post-spawn stage of the female of *E. vallentini* collected. The same probably applies for the changes in PE recorded for *T. macrura* females.

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Key words: Antarctic, phospholipids, triacylglycerols, trophic status, wax esters, zooplankton

Introduction

Eleven species of euphausiids inhabit the Southern Ocean but only six of these are important and endemic south of the Antarctic Polar Front (Knox 1994). *Euphausia superba* Dana is the dominant species within of the seasonal ice zone (SIZ) and shares to various extents this oceanic domain with other species, such as *Thysanoessa macrura* Sars and *Euphausia frigida* Hansen. North and south of these limits other species are of increasing importance, e.g. *Euphausia crystallorophias* Holt & Tattersall near the Antarctic coastline, and north of the Antarctic Polar Front *Euphausia vallentini* Stebbing and *E. triacantha* Holt & Tattersall. Because their lipid dynamics and life cycles are tightly linked, they should show different strategies of accumulation and metabolism with changes in both environment (changes in trophic interactions) and reproductive strategies.

Over the past two decades various works have clarified some of the main features of lipid structure and metabolism of *E. superba* (Virtue *et al.* 1993, Mayzaud *et al.* 1998, Cripps *et al.* 1999). The other euphausiid species have received far less attention (Falk-Petersen *et al.* 1999, 2000) although they all play an important role in the dynamic of the Antarctic food web and support large populations of higher predators (Mauchline & Fisher 1969, Ridoux 1988). Recently, the lipid metabolism of *T. macrura* has received some attention because of its unique wax ester composition (Kattner *et al.* 1996, Hagen & Kattner 1998, Falk-Petersen *et al.* 1999). Ontogenic changes of total fatty acids and alcohols as well as the influence of sex on the major lipid classes have been reported. However, further information is needed on the younger immature stages as well as the role of polar lipids with the possible accumulation of phosphatidylcholine (Hagen *et al.* 1996, Mayzaud 1997). Our knowledge of the life history and lipid dynamics of *E. vallentini* remains largely unknown, with only a single report by Attwood & Hearshaw (1992).

The objectives of the present research were:

1) to determine the structure of the main polar lipid as well as neutral lipid fractions with respect to sex (*E. vallentini*), growth stage and location (*T. macrura*), and

2) to compare the trophic position of these two species

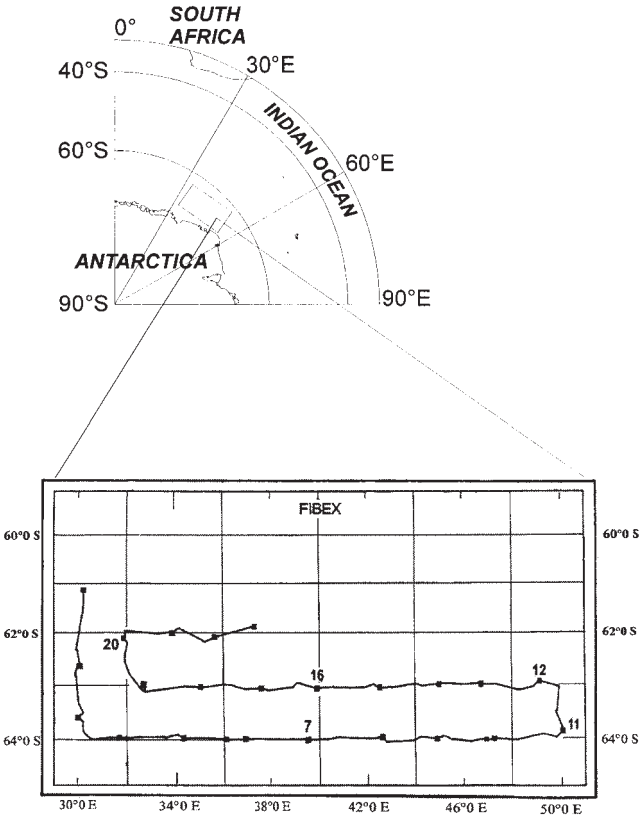

Fig. 1. Sampling stations for *Thysanoessa macrura*.

with the other Antarctic species using lipid biomarkers.

Material and methods

Sampling and size measurement

Thysanoessa macrura was obtained from RMT 8 oblique tows made to a depth of 100 m during a cruise of the RV *Marion Dufresne* in February 1981 at five stations located in the Indian sector of the Southern Ocean (Fig. 1). *Euphausia vallentini* were sampled in January 1998 on the Kerguelen continental shelf during a cruise of the RV *La Curieuse* at a single station at 48°30'S, 70°30'E. All samples were sorted immediately after capture, rinsed with distilled water and deep frozen (-80°C). They were stored at -80°C under nitrogen until analysis.

Subsets of 10–15 *T. macrura* were identified individually to development or sexual stage, measured (Body Length or BL: Standard Length 1; Mauchline 1980), weighed (wet weight: WW) and extracted for lipid class analyses. Juveniles and females, which dominated at the two stations were identified, pooled and extracted to obtain sufficient material for fatty acid determination in the main lipid classes. Similarly, groups of 40–45 male and female *E. vallentini* were sorted individually, measured (BL: Standard Length 1), weighed (wet weight: WW) and extracted for lipid class analyses. The lipid extracts were then pooled to obtain three replicated samples for fatty acid determination.

Lipid extraction and determination

Extraction and analyses took place immediately upon return to the laboratory. Entire krill was placed frozen on crushed ice and brought to 0°C. Size and wet weight were measured before lipid extraction according to the method of Bligh & Dyer (1959). After solvent evaporation at high vacuum, the extracted lipids were weighed in tared vials and the lipid extracts were placed in nitrogen at -70°C until analysis.

Lipid classes were quantified after chromatographic separation coupled with FID detection on an Iatroscan Mark III TH 10 for *T. macrura* and Mark IV for *E. vallentini* (Ackman 1981). Total lipid extracts were applied to Chromarods SII (*T.m*) and SIII (*E.v*) using microcapillaries (1 µl) and analysed in duplicate. Neutral lipids were separated using a double development procedure with the following solvent systems: *n*-hexane: benzene: formic acid 80:20:1 (v/v) followed by *n*-hexane: diethylether: formic acid 97:3:1.5 (v/v). Phospholipids were separated with chloroform: methanol: NH_4 50:50:5 (v/v). All listed lipid classes, and in particular free fatty acids, were separated and identified. Calibration was achieved using commercial standards, except for polar lipids (phosphatidylcholine and phosphatidylethanolamine), wax esters and triacylglycerols, which were purified from both species by column chromatography and TLC (Mayzaud *et al.* 1998) and used as standards.

Fatty acid methyl esters of total lipids were prepared with 7% boron trifluoride in methanol (Morrison & Smith 1964). Fatty alcohols were acetylated using acetic anhydride. Gas liquid chromatography (GLC) of all esters was carried out on a 30 m length x 0.32 mm internal diameter quartz capillary column coated with Famewax (Restek) in a Perkin-Elmer XL Autolab gas chromatograph equipped with a flame ionization detector (FID). The column was operated isothermally at 190°C for methyl esters and 200°C for alcohol acetates. Helium was used as carrier gas at 7 psig. Injector and detector were maintained at 250°C. Individual components were identified by comparing retention time data with those obtained from authentic and laboratory standards. In addition to the examination of esters as recovered, a part of all ester samples was completely hydrogenated and the products examined qualitatively and quantitatively by GLC. The level of accuracy is ± 5% for major components, 1–9% for intermediate components and up to ± 30% for minor components.

Statistical treatment

The allometric relationships between dry or wet weight (DW or WW) and length (BL) for the different maturity

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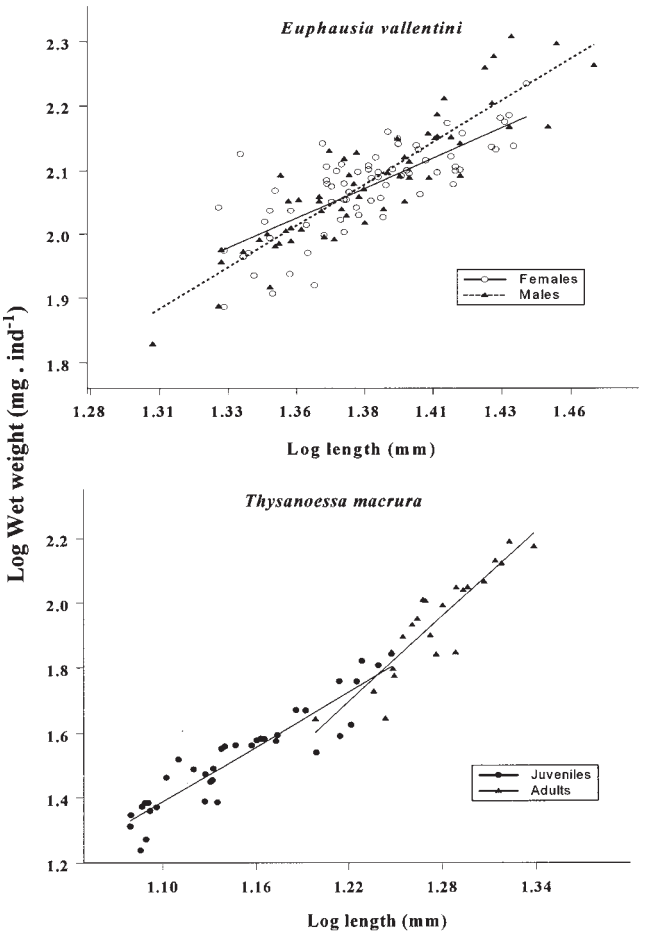

Fig. 2. Log-log regressions between wet weight and length for juveniles and adult stages of *T. macrura*, and males and females of *E. vallentini*.

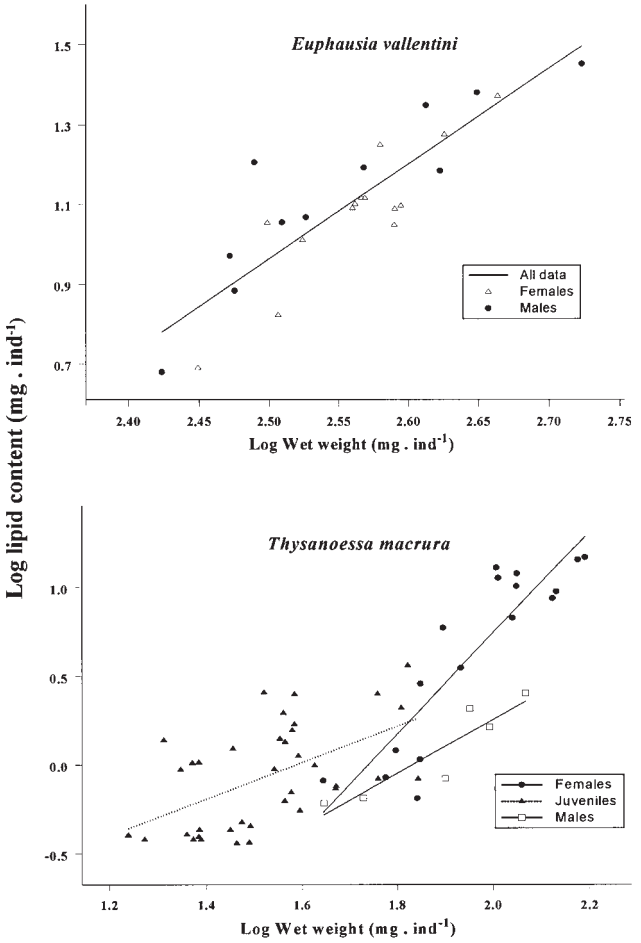

Fig. 3. Log-log relationships between wet weight and total lipid content for male and female stages of *E. vallentini* and juveniles, males and females of *T. macrura*.

stages (WW = a * BL^b) was computed after log-log transformation and model I regression (Sokal & Rohlf 1981). Multiple comparisons were made using an ANOVA followed by Tukey *post hoc* tests. Percentage data were normalized using arcsine transformation (Zar 1984). Bivariate analyses were made with SYSTAT 10.

Differences and classification of fatty acid profiles was achieved by multiple discriminant analysis (Cooley & Lohnes 1971). Briefly, a discriminant reference model was established using the literature data on the fatty acid composition of total lipids from Antarctic euphausiids. The model derives the component which best separates the group of observations in the measurement space. Normality is presupposed and although discriminant analysis is known to be fairly robust, transformation of percentage data was needed. Computation was made using SIMCA 9.0 software, which has developed a modification that allows new observations with an unknown group membership in the current model to be to classified and tested. Fatty acid data from *T. macrura* and *E. vallentini* were used as supplementary observations. Classification of polar lipids was achieved by clustering the fatty acid composition using the Bray and Curtis distance index and unweighed pair group average linkage (Pielou 1984). Computation was made using MVSP 3.1.

Results

Size, weight and lipid relationships

The log-linear relationships between wet weight and length were established for all stages present in the samples collected, i.e. juvenile (when present) male and female. Because of the small number of *T. macrura* males collected (n = 7), no significant differences were observed between the weight-length regressions for male and female ($P > 0.05$). Hence, the data were pooled as adult stages. The regressions for juveniles and adults (Fig. 2) were both highly significant and significantly different (slope test $F_{1,56} = 11.26$; $P = 0.001$), corresponding to two general equations:

Adults: Log WW = 4.38 log BL-3.64 ($r^2 = 0.814$; $F_{1,21} = 91.9$, $P = 0.0001$)

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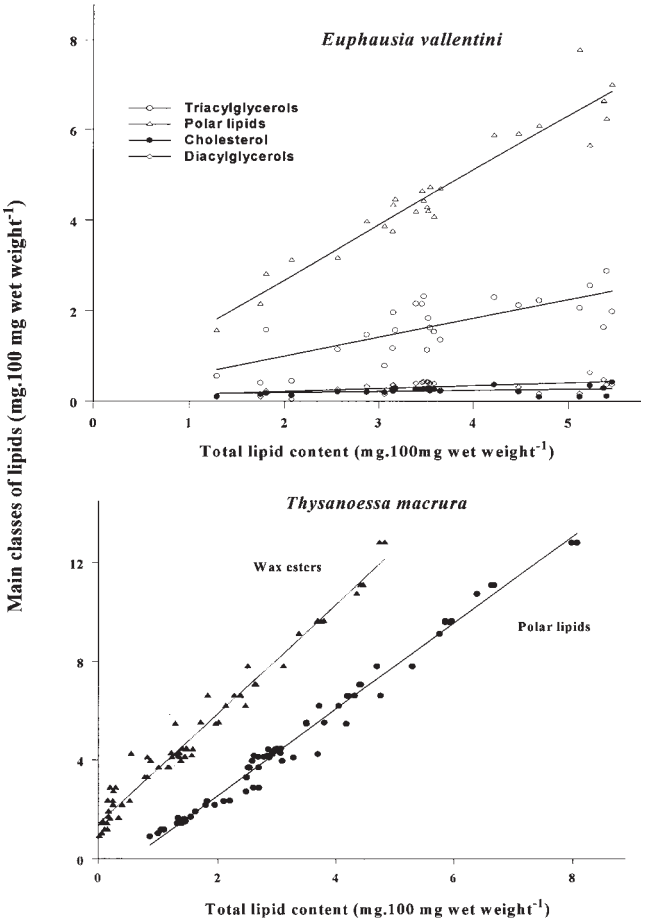

Fig. 4. Relationships between total lipid content and concentration of the main lipid classes in whole krill of *E. vallentini* and *T. macrura*. All developmental stages sampled at the various location were combined.

Juveniles: Log WW = 2.83 log BL-1.72 ($r^2 = 0.859$; $F_{1,35} = 214$, $P = 0.0001$)

Similarly, *E. vallentini* showed significant log-linear regressions between length and weight (Fig. 2) for male and female, both significantly different (slope test $F_{1,124} = 7.92$; $P = 0.006$) with the following corresponding equation:

Males: Log WW = 2.60 log BL-1.53 ($r^2 = 0.804$; $F_{1,55} = 230$, $P = 0.0001$)

Females: Log WW = 1.87 log BL-0.52 ($r^2 = 0.575$; $F_{1,69} = 93.3$, $P = 0.0001$)

Total lipid content was also correlated with the changes in wet weight for both species (Fig. 3). However, while log-linear regressions for *E. vallentini* failed to show significant differences between sexes ($P > 0.05$), *T. macrura* showed significantly different log-linear regressions with sex and developmental stage (slope tests male/female: $F_{1,19} = 4.54$, $P = 0.04$; slope test juvenile/female: $F_{1,40} = 30.3$, $P = 0.0001$; slope test juvenile/male: $F_{1,50} = 0.012$, $P = 0.91$and test for sample means: $F_{1,51} = 27.5$, $P = 0.0001$). Hence, data for both sexes of *E. vallentini* were pooled in a single regression: log Lipt = 2.39 logWW-5.01 ($r^2 = 0.757$; $F_{1,23} = 71.7$, $P = 0.001$). Regression equations for *T. macrura* showed a decreasing allometric exponent from female: log Lipt = 2.86 logWW-4.97 ($r^2 = 0.792$; $F_{1,15} = 57.3$, $P = 0.001$), to male: log Lipt = 1.53 logWW-2.80 ($r^2 = 0.835$; $F_{1,4} = 20.3$, $P = 0.01$) and juvenile: log Lipt = 1.04 logWW-1.65 ($r^2 = 0.300$; $F_{1,35} = 14.8$, $P = 0.001$), suggesting a faster rate of lipid accumulation in females and somewhat similar rate in juveniles and males.

In both species, changes in total lipids were linearly

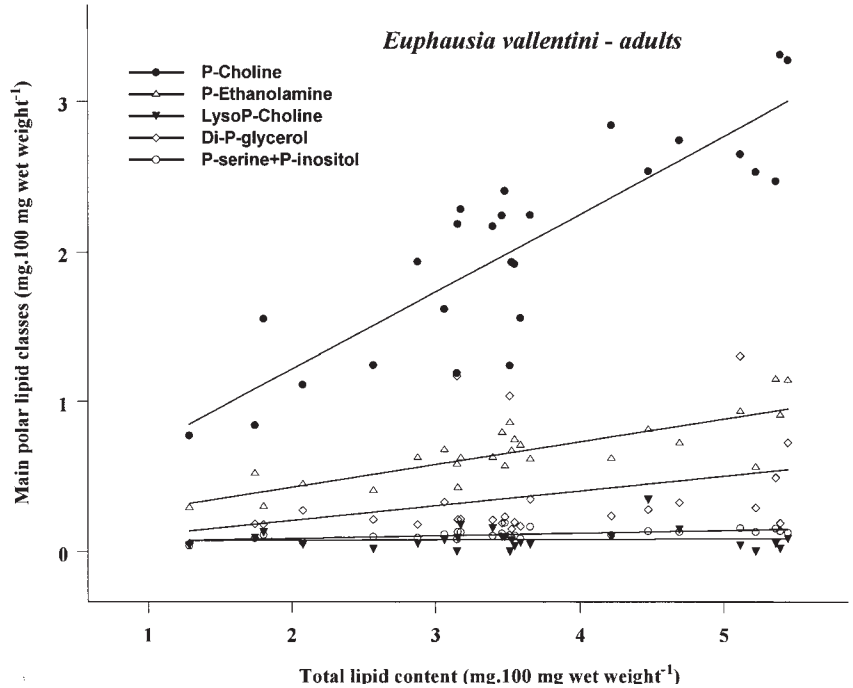

Fig. 5. Relationships between total lipids content and the concentrations of the main polar lipids in adult stages of *E. vallentini*.

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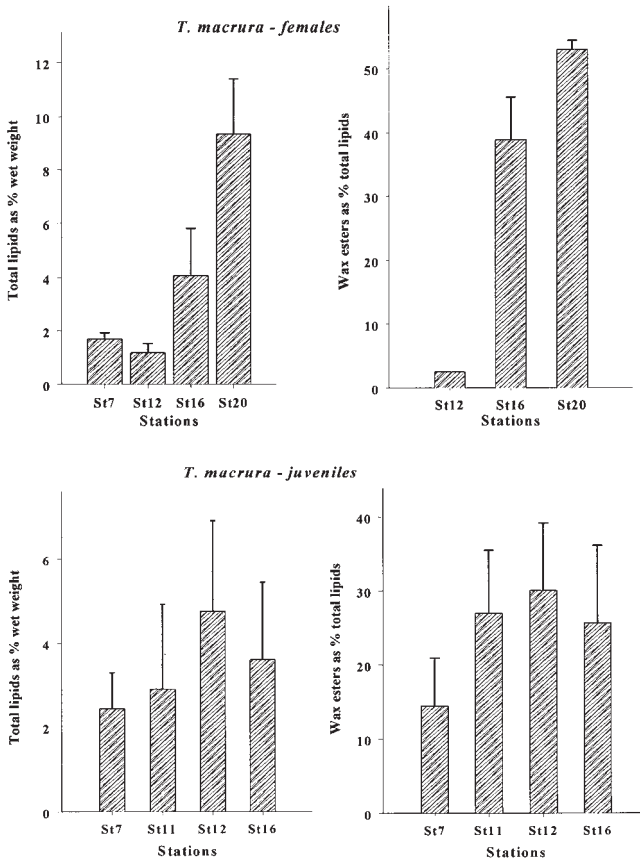

Fig. 6. Spatial changes in total lipid and wax esters content in *T. macrura* females and juveniles (see Fig. 1 for position of station number).

related to structural polar lipids and reserve lipids (Fig. 4). Interestingly, the data suggest different controls in the dynamics of lipid changes between the two species. Wax esters dominated the changes in *T. macrura* while polar lipid explained the largest part of the variation observed in *E. vallentini*. In this latter case, diacylglycerols and cholesterol remained constant throughout the range of total lipid content. Within the polar lipid fraction, the detailed analysis with *E. vallentini* showed (Fig. 5) that phosphatidylcholine (PC) and phosphatidylethanolamine (PE) were significantly correlated with the changes in total lipid content, contrary to the diphosphoglycerol (DPG), phosphatidylserine+inositol (PS-PI) and lysophosphatidylcholine (LPC) fractions. The regression equations for the PC (PC = 0.52 Lip tot; $r^2 = 0.742$; $F_{1,24} = 68.8$, $P = 0.0001$) and PE (PE = 0.15 Lip tot; $r^2 = 0.656$; $F_{1,24} = 45.8$, $P = 0.0001$) confirmed the major contribution of PC to the dynamic of lipid changes.

Multiple comparisons, using ANOVA, to test differences in total lipid content in *T. macrura* between sampling locations showed that females and juveniles had significantly higher lipid content at station 20 and 12 respectively (Fig. 6; $F_{3,13} = 14.26$, $P = 0.0001$ for females; $F_{4,32} = 6.11$, $P = 0.001$ for juveniles). Similar ANOVA between sex and developmental stages showed significant differences in wax esters and polar lipids with concentrations ranking females > juveniles > males ($F_{2,65} = 23.68$, $P = 0.0001$). Wax esters concentrations followed the pattern observed for total lipids with higher values at station 20 and 12 and minimum values at station 12 and 7 for females and juveniles respectively (Fig. 6). The same test with *E. vallentini* failed to show differences in concentrations of triacylglycerols, polar lipids and most polar lipid constituents (PC, PE, DPG). Significant differences were observed only for cholesterol (male > female; $F_{1,76} = 5.20$, $P = 0.02$), diacylglycerols (male > female; $F_{1,76} = 7.48$, $P = 0.007$) and PS-PI (male > female; $F_{1,74} = 5.15$, $P = 0.02$).

Lipid composition in relation to sex and developmental stage

Detailed compositions of the lipids from species, sexes and developmental stages are presented in Table I. Detailed lipid

Table I. Lipid content and composition in lipid classes of *Thysanoessa macrura* and *Euphausia vallentini* in relation to sex and maturity stage.

	Thysanoessa macrura			*Euphausia vallentini*	
	Juveniles $n = 37$	Males $n = 6$	Females $n = 17$	Females $n = 14$	Males $n = 12$
Total lipids (% wet weight)	3.02 ± 1.7	1.64 ± 0.53	9.36 ± 2.0	3.42 ± 0.9	3.76 ± 1.4
Lipid classes (% total lipids)	Station 12		Station 20		
Phosphatidylcholine	33.9 ± 3.8	—	45.1 ± 0.2	47.5 ± 6.3	45.5 ± 6.7
Phosphatidylethanolamine	26.5 ± 1.1	—	0.3 ± 0.04	9.1 ± 2.3	11.2 ± 3.0
Phophatidylserine + inositol	ND	—	ND	2.7 ± 0.6	2.7 ± 0.6
Diphosphatidylglycerol	ND	—	ND	5.8 ± 5.3	4.9 ± 4.0
Lysophosphatidyl choline	ND	—	ND	2.4 ± 1.6	1.3 ± 0.9
Diacylglycerols	ND	—	ND	4.3 ± 1.8	5.0 ± 1.1
Triacylglycerols	2.3 ± 0.3	—	ND	22.3 ± 5.3	31.9 ± 4.6
Free fatty acids	2.7 ± 1.9	—	1.6 ± 1.1	0.9 ± 0.8	0.8 ± 0.8
Cholesterol	3.8 ± 0.1	—	ND	3.5 ± 1.2	3.7 ± 0.9
Wax esters	30.8 ± 3.6	—	53.0 ± 0.6	ND	ND

Symbols: ND = not detected, — = not analysed, ± SD = standard deviation

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Table II. Fatty acid composition of the main lipid classes of female and juvenile *Thysanoessa macrura*.

Fatty acids	Juveniles (% total fatty acids or alcohols)						Females (% total fatty acids or alcohols)				
	Total	TG	WE-Fa	WE-Alc	PC	PE	Total	WE-Fa	WE-Alc	PC	PE
14:0	17.3	25.3	55.5	0.4	4.7	2.4	27.1	47.7	0.2	5.6	2.1
Iso15:0	0.5	0.6	1.7	-	0.2	-	0.4	0.5	0.1	0.2	-
Aiso15:0	-	0.5	-	-	-	-	-	0.2	-	-	-
15:0	0.7	0.9	1.3	-	0.6	0.2	0.5	0.5	< 0.1	0.3	0.2
Iso 16:0	-	0.4	-	-	0.2	0.2	0.2	-	-	0.2	0.3
16:0	21.7	25.2	18.1	1.6	27.7	16.5	26.9	21.6	0.8	31.4	30.7
17:0	-	-	0.4	-	0.3	-	0.3	0.4	< 0.1	0.3	0.1
Iso 18:0	-	-	-	-	0.2	-	-	0.2	< 0.1	0.3	0.3
18:0	0.6	1.5	0.5	1.4	0.5	0.8	0.8	0.6	1.1	0.7	1.6
Σ Saturates	40.0	53.4	76.3	3.3	33.5	19.8	55.6	71.2	2.2	38.6	35.1
16:1n-9	0.5	0.8	0.9	-	0.4	0.2	0.6	0.8	-	0.4	0.2
16:1n-7	2.6	9.3	3.3	-	2.5	0.8	3.2	3.2	0.3	2.6	1.2
16:1n-5	-	1.5	3.0	-	0.5	-	1.2	2.1	0.1	0.5	0.4
18:1n-9	7.5	11.9	7.3	30.5	7.9	4.9	10.0	10.2	34.4	7.7	10.8
18:1n-7	5.1	4.9	5.0	48.1	3.6	9.2	4.2	4.4	29.5	2.6	12.5
18:1n-5	-	-	0.4	-	0.3	0.4	0.7	0.7	1.1	0.7	1.1
20:1n-9	-	0.5	0.5	15.3	0.2	0.4	1.0	1.5	22.3	-	0.8
20:1n-7	-	-	-	0.4	-	-	0.1	0.3	1.4	-	-
22:1n-11+13	-	-	-	0.5	-	-	0.2	0.4	3.2	-	-
22:1n-9	-	-	-	0.7	-	-	0.3	0.2	1.9	-	-
24:1	-	-	-	-	-	-	0.9	0.2	0.2	1.4	0.3
Σ monoenes	15.8	28.9	20.4	94.2	15.3	15.8	22.3	23.6	94.4	15.9	27.3
18:2n-6	4.0	3.3	0.6	1.4	4.1	2.3	1.7	0.5	0.7	2.8	2.2
16:3n-4	-	1.2	-	-	0.1	-	-	-	-	-	-
18:3n-6	-	-	-	-	0.4	-	0.2	-	-	0.3	-
18:3n-3	0.8	0.6	-	-	0.6	0.5	0.3	-	< 0.1	0.6	0.6
20:3n-6	-	-	-	-	0.5	-	0.2	-	-	0.3	-
Σ poly with 3	0.8	1.8	-	-	1.6	0.5	0.7	-	< 0.1	1.3	0.6
16:4n-3	-	-	-	-	0.3	-	0.4	-	-	-	-
18:4n-3	0.5	0.9	0.7	-	0.8	-	0.7	0.5	0.1	1.2	0.9
20:4n-6	2.1	-	-	-	1.5	1.5	0.6	-	-	1.1	-
20:4n-3	-	-	-	-	0.7	0.4	0.3	-	-	0.6	-
Σ poly with 4	2.6	0.9	0.7	-	3.3	1.9	2.0	0.5	0.1	2.9	0.9
20:5n-3	17.3	3.7	0.5	-	23.9	16.7	9.8	1.2	0.6	24.6	12.4
22:6n-3	17.6	5.7	0.2	-	15.7	40.5	5.6	0.3	0.1	12.1	20.9

TG = triacylglycerols, WE-Fa = wax esters fatty acids, WE-Alc = wax esters alcohols, PC = phosphatidylcholine, PE = phosphatidylethanolamine.

class composition (neutral and polar) could only be established for individuals collected at station 20 for females and station 12 for juveniles. Total lipid concentrations (% wet weight) varied from a mean value of 1.64% to 9.36%, in male and female of *T. macrura* respectively. Intermediate values around 3.4% were recorded for juvenile of *T. macrura* and both sexes of *E. vallentini*. In terms of percentage of total lipids, neutral lipids were dominated by wax esters in *T. macrura* and triacylglycerols in *E. vallentini*, regardless of sex or stage. Interestingly, triacylglycerols were present in juvenile stages of *T. macrura* but were below detection level in adult females. Free fatty acids were low in all cases with values ranging from less than 1% in *E. vallentini* to 2.7% in juveniles of *T. macrura*. Monoacylglycerols were not detected and diacylglycerols were present at intermediate levels in both sexes of *E. vallentini*. Polar lipids (% total lipids) were dominated by phosphatidylcholine followed by phosphatidylethanolamine in all cases except one. Indeed, *T. macrura* females showed extremely low levels of PE compared to juvenile stages. Because of the larger sample size, a more detailed analysis of the polar fraction was achieved for *E. vallentini* with significant percentages of diphosphoglycerol and to a lesser extent of phospatidylserine + inositol and lysophosphatidylcholine.

Multiple comparisons with ANOVA showed significant differences in wax esters and polar lipids with developmental stage in *T. macrura*, with higher percentages of wax esters and lower percentages of polar lipids in females than in juveniles ($F_{1,54}$ = 11.63, P = 0.0001). No differences were observed between males and females in *E. vallentini* for any of the lipid classes considered.

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Table III. Fatty acid composition of total polar lipids and the main class of neutral lipids of *Euphausia vallentini* male and female ($n = 5$).

Fatty acid	Male (% total fatty acids)				Female (% total fatty acids)			
	Total	TG	DG	PL	Total	TG	DG	PL
14:0	6.2 ± 0.5	11.5	2.1	0.6	5.4 ± 0.2	9.9	2.3	0.7
Iso 15:0	0.2 ± 0.01	0.4	0.1	< 0.1	0.2 ± 0.01	0.3	0.1	< 0.1
Anteiso 15:0	0.1 ± 0.01	0.1	< 0.1	< 0.1	0.1 ± 0.01	0.1	0.1	< 0.1
15:0	0.3 ± 0.01	0.6	0.2	0.1	0.3 ± 0.01	0.5	0.2	0.1
Iso 16:0	0.1 ± 0.01	0.2	0.1	0.1	0.1 ± 0.0	0.1	0.1	0.1
Iso 17:0	0.3 ± 0.01	0.4	0.3	0.3	0.3 ± 0.01	0.4	0.3	0.3
16:0	25.8 ± 0.6	28.9	16.4	22.2	25.6 ± 0.5	29.1	18.5	23.2
Phytanic acid	1.1 ± 0.03	2.1	0.8	0.1	1.2 ± 0.02	2.1	1.3	0.1
Iso 18:0	0.2 ± 0.01	0.2	0.3	0.2	0.2 ± 0.02	0.2	0.3	0.2
18:0	0.9 ± 0.03	1.2	1.1	0.9	0.9 ± 0.03	1.0	1.5	0.9
24:0	0.2 ± 0.02	< 0.1	0.4	< 0.1	0.2 ± 0.01	< 0.1	0.4	0.1
Σ saturates	35.6 ± 1.1	46.1	22.3	22.3	34.7 ± 0.6	44.1	25.4	25.9
14:1	0.1 ± 0.01	0.2	< 0.1	-	0.1 ± 0.01	0.2	0.1	-
16:1n-7	9.1 ± 0.3	17.7	5.9	1.8	8.5 ± 0.2	17.0	6.1	2.1
16:1n-5	0.3 ± 0.02	0.5	0.3	0.1	0.3 ± 0.0	0.5	0.3	0.1
17:1	0.4 ± 0.0	0.5	0.4	0.2	0.4 ± 0.01	0.6	0.5	0.2
18:1n-9	10. ± 0.21	14.9	9.7	7.2	10. ± 0.14	14.6	11.5	8.1
18:1n-7	5.0 ± 0.1	5.7	5.7	4.4	5.1 ± 0.1	5.6	6.6	4.6
18:1n-5	0.3 ± 0.0	0.3	0.3	0.3	0.3 ± 0.02	0.4	0.5	0.3
20:1n-9	1.3 ± 0.0	1.7	1.3	1.2	1.3 ± 0.05	1.8	1.7	1.1
20:1n-7	0.1 ± 0.0	0.2	0.1	0.1	0.2 ± 0.02	0.2	0.1	0.1
22:1n-11+13	-	< 0.1	-	-	-	0.1	0.1	-
22:1n-9	0.5 ± 0.0	0.2	0.3	1.0	0.4 ± 0.02	0.2	0.3	0.8
22:1n-7	0.1 ± 0.0	< 0.1	0.1	0.2	0.1 ± 0.02	0.1	0.1	0.2
24:1	0.7 ± 0.1	<0.1	0.1	1.7	0.6 ± 0.02	< 0.1	0.1	1.4
Σ monoenes	28. 1 ± 0.2	41.9	24.1	18.2	27.7 ± 0.1	41.1	27.8	19.1
16:2n-4	1.0 ± 0.01	1.9	0.5	0.1	0.9 ± 0.02	1.9	0.5	0.1
18:2n-6	1.2 ± 0.02	0.9	1.3	1.5	1.1 ± 0.03	0.8	1.2	1.4
20:2n-6	0.1 ± 0.01	0.1	0.1	0.1	0.1 ± 0.01	0.1	0.1	0.1
22:2n-6	0.1 ± 0.04	-	< 0.1	0.1	0.1 ± 0.02	-	< 0.1	0.1
Σ dienes	2.5 ± 0.1	2.9	2.1	1.8	2.2 ± 0.04	2.9	1.9	1.7
16:3n-4	0.1 ± 0.0	0.2	-	-	0.1 ± 0.00	0.3	-	-
18:3n-6	0.1 ± 0.02	0.1	0.1	0.1	0.1 ± 0.04	0.1	0.1	0.1
18:3n-3	0.5 ± 0.02	0.5	0.5	0.5	0.5 ± 0.03	0.5	0.5	0.5
Σ trienes	0.7 ± 0.04	0.9	0.6	0.6	0.7 ± 0.07	0.9	0.6	0.6
16:4n-1	0.6 ± 0.01	0.8	0.3	-	0.6 ± 0.01	1.1	0.3	-
18:4n-3	1.7 ± 0.04	2.1	1.3	0.8	1.9 ± 0.04	2.9	1.4	0.9
18:4n-1	0.1 ± 0.01	0.1	0.1	-	0.1 ± 0.03	0.1	0.1	-
20:4n-6	0.5 ± 0.03	< 0.1	0.7	0.7	0.5 ± 0.03	< 0.1	0.6	0.7
20:4n-3	0.4 ± 0.02	0.2	0.6	0.5	0.4 ± 0.01	0.2	0.5	0.5
Σ poly with 4	3.3 ± 0.1	3.2	3.0	2.1	3.4 ± 0.10	4.4	2.9	2.1
20:5n-3	18.6 ± 0.5	2.6	29.4	32.3	19.1 ± 0.3	3.9	25.7	30.3
21:5n-3	0.5 ± 0.04	0.2	0.4	0.8	0.6 ± 0.03	0.2	0.5	0.9
22:5n-3	0.3 ± 0.03	0.2	0.4	0.5	0.4 ± 0.01	0.2	0.4	0.6
Σ poly with 5	19.5 ± 0.5	3.0	30.3	33.6	20.0 ± 0.3	4.3	26.7	31.8
22:6n-3	6.7 ± 0.4	1.5	17.1	18.3	10.7 ± 0.3	1.8	14.3	18.4

Symbols: TG = triacylglycerols, DG = diacylglycerols, PL = polar lipids.

Fatty acids and alcohols constituents of T. macrura *in relation to developmental stage*

The relative composition of total fatty acids and fatty alcohols is presented in Table II for both juvenile and female stages and the major lipid classes. Fatty acid composition of the total lipids was driven by the abundance of wax esters. In both stages, the saturated acids palmitic (16:0) and myristic (14:0) dominate. Highly unsaturated fatty acids (HUFA): eicosapentaenoic acid (EPA 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3) were also largely represented but with higher percentages in juveniles than in females. Monoenoic acids were accounted for mostly by oleic (18:1n-9) and vaccenic (18:1n-7) acids and to a minor extent by palmitoleic acid (16:1n-7). The ratio

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18:1n-9/18:n-7 was minimum in juveniles (1.47 and 2.36 respectively in juveniles and females) because of a higher proportion of oleic acid in wax ester rich females. Dienoic and trienoic acids showed similar proportion in both cases while polyenes with 4 double bonds showed larger concentrations of 20:4n-6 in juveniles than in females.

Wax esters displayed relatively similar composition for both juvenile and female stages (Table II): myristic acid (14:0) dominated followed by palmitic acid (16:0), oleic acid (18:1n-9) and vaccenic acid (18:1n-7). Polyunsaturated acids showed low percentages with less than 2% of EPA and DHA. Similarly oleyl, vaccenyl, 20:1n-9 and 22:1n-11 alcohols accounted for 94% of total alcohols in both stages. However, higher percentages of myristic acid and lower percentages of palmitic acid, oleic acid and EPA were recorded in juvenile. As a result, the ratio 18:1n-9/18:1n-7 was lower in juveniles (1.4) than in females (2.3). In terms of fatty alcohols, juveniles showed higher percentages of 16:0, 18:1n-7 and 18:2n-6, and lower percentages of 18:1n-9, 20:1n-9, 22:1n-13, 22:1n-11 and EPA than females. Again the ratio between 18:1n-9/18:1n-7 alcohols was lower in juveniles (0.6) than in females (1.2). The rather specific pattern of fatty alcohols is in perfect agreement with the synthetic pathway described by Kattner *et al*. (1996).

Two classes of polar lipids were analysed for the fatty acid constituents: PC and PE (Table II). PC showed relatively similar composition in both juveniles and females with the dominance in decreasing order of palmitic acid, EPA and DHA. Oleic, vaccenic and linoleic (18:2n-6) were also present with percentages exceeding 3–4%. On the contrary, PE showed different patterns of fatty acid composition : females presented far higher percentages of palmitic and oleic acids and to a lesser extent of stearic, palmitoleic, vaccenic and 18:4n-3 than juveniles. Conversely, lower concentrations in females than in juveniles of EPA, DHA and arachidonic acids were recorded.

Lipid classes constituents of E. vallentini *in relation to sex*

The fatty acid relative composition of neutral lipid classes and total polar lipids from male and female *E. vallentini* showed a high degree of similarity (Table III). Standard deviations of three GLC analyses are indicated for total lipids but apply to all other lipid classes and confirmed the accuracy indicated in the method section. Triacylglycerols were in both cases dominated by saturated and monoenoic acids with palmitic, palmitoleic, oleic and myristic acids accounting for more than 70% of total fatty acids. Similarly, polyunsaturated acids were represented mostly by 18:4n-3, EPA, DHA and 16:2n-4. Phytoplankton descriptors such as C16-polyunsaturated acids were present with slightly higher percentages in females. As usual, total polar lipids showed a high degree of polyunsaturation with EPA and DHA as

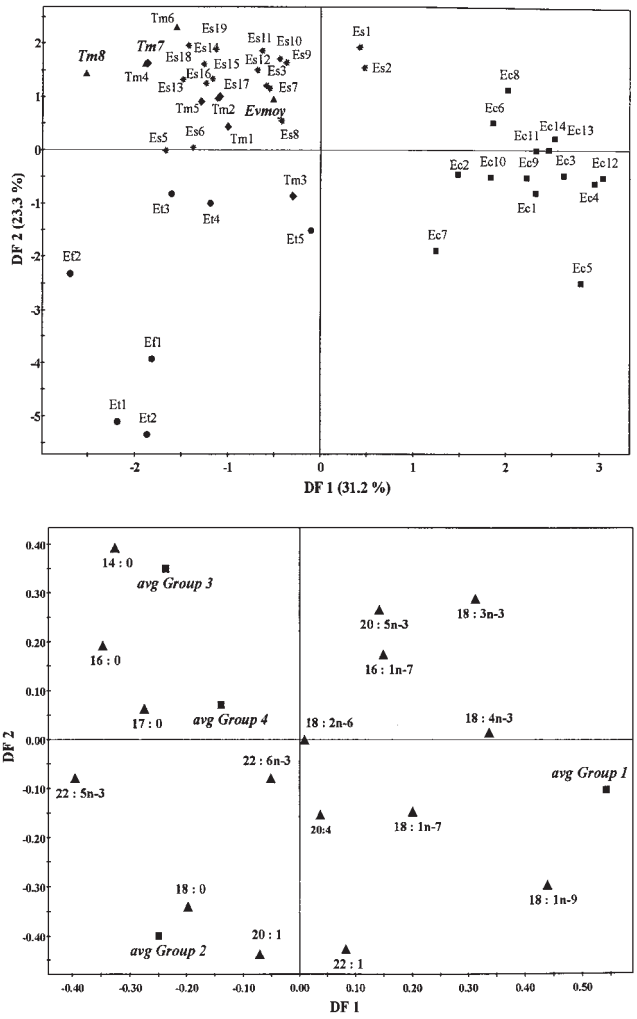

Fig. 7. PLS-discriminant analysis of Antarctic euphausiids based on their total lipid fatty acid structure. Plot of scores (top) for the first two discriminant functions based on published data as active observations: Ec = *E. crystallorophias* (Kattner & Hagen 1998), Ef = *E. frigida* (Phleger *et al.* 1998, 2002), Et = *E. triancantha* (Cripps & Atkinson 2000, Phleger *et al.* 2002), Es = *E. superba* (Bottino 1975, Phleger *et al.* 1998, 2002, Mayzaud 1997), *T. macrura* (Kattner *et al.* 1996, 1998). Present data (bold and italic) were added as illustrative observations. Plot of loadings or fatty acids (bottom) for the same functions with the position of the average scores of the four groups of species.

major constituents in both sexes. Palmitic exceeded 20% of total acids and oleic, palmitoleic as well as linoleic acids complement the fatty acid structure. Diacylglycerols showed a composition intermediate between neutral and polar lipids with relatively high levels of EPA and DHA, presence of C16-polyunsaturated acids but with percentages of palmitoleic acid, myristic acid, oleic acid and 18:4n-3 higher than in the total polar lipid fraction, and levels of palmitic acid lower than in polar lipids and triacylglycerols.

Detailed fatty acid composition of the main polar lipid

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Table IV. Fatty acid composition of the main polar lipid fractions from male and female of *Euphausia vallentini*.

Fatty acids	Male			Female		
	PC	PE	LPC	PC	PE	LPC
14:0	0.6 ± 0.1	1.0	1.9	1.0 ± 0.1	1.0	0.3
Iso 15:0	0.1 ± 0.01	0.3	1.6	0.1 ± 0.00	0.4	0.1
Anteiso 15:0	< 0. ± 0.01	0.1	0.2	< 0. ± 0.01	0.2	< 0.1
15:0	0.2 ± 0.02	0.3	0.4	0.2 ± 0.01	0.1	0.1
Iso 16:0	0.1 ± 0.01	0.4	1.1	0.1 ± 0.00	0.1	0.2
Iso 17:0	0.4 ± 0.01	0.6	0.9	0.3 ± 0.00	0.4	0.4
Anteiso 17:0	0.1 ± 0.01	0.2	0.6	0.1 ± 0.00	0.1	0.1
16:0	34. ± 0.45	21.7	16.0	32. ± 0.7	14.5	9.5
17:0	0.1 ± 0.0	0.3	0.8	0.1 ± 0.01	0.2	0.3
Phytanic acid	0.1 ± 0.0	< 0.1	0.2	0.1 ± 0.00	< 0.1	0.2
Iso 18:0	0.3 ± 0.01	0.6	1.2	0.2 ± 0.03	0.5	0.4
18:0	1.2 ± 0.04	3.8	15.3	0.9 ± 0.02	2.3	4.4
22:0	0.1 ± 0.01	0.2	0.9	0.1 ± 0.01	0.1	0.3
24:0	0.1 ± 0.01	0.3	-	0.1 ± 0.01	0.3	-
Σ saturates	37. ± 0.6	29.7	41.0	35.4 ± 0.8	20.4	16.4
16:1n-7	2.6 ± 0.1	0.9	1.7	2.8 ± 0.1	0.9	1.6
16:1n-5	0.1 ± 0.01	0.2	1.2	0.1 ± 0.01	0.2	0.3
17:1	0.3 ± 0.01	0.3	0.5	0.2 ± 0.01	0.2	0.5
18:1n-9	10. ± 0.19	6.6	10.6	10. ± 0.040	6.3	30.3
18:1n-7	5.1 ± 0.04	9.3	3.6	4.4 ± 0.02	8.8	7.3
18:1n-5	0.3 ± 0.01	0.5	0.3	0.3 ± 0.01	0.6	0.3
20:1n-9	1.8 ± 0.02	1.1	1.8	1.3 ± 0.02	1.1	2.0
20:1n-7	0.2 ± 0.01	0.2	0.4	0.1 ± 0.01	0.2	0.2
22:1n-9	1.7 ± 0.05	0.1	1.1	1.1 ± 0.03	0.2	0.5
22:1n-7	0.4 ± 0.02	-	0.2	0.3 ± 0.02	-	0.2
24:1	2.9 ± 0.2	0.1	1.8	1.9 ± 0.05	0.2	1.2
Σ monoenes	26. ± 0.1	19.2	23.2	22.5 ± 0.09	18.6	44.2
16:2n-4	0.1 ± 0.01	0.2	0.4	0.1 ± 0.01	0.1	0.1
18:2n-9	0.1 ± 0.01	0.2	0.4	0.1 ± 0.01	0.2	0.1
18:2n-6	1.9 ± 0.01	0.6	1.1	1.4 ± 0.02	1.1	2.4
20:2n-6	0.1 ± 0.01	0.1	0.1	0.1 ± 0.0	0.1	0.2
Σ dienes	2.3 ± 0.01	1.1	1.9	1.8 ± 0.02	1.5	2.8
18:3n-3	0.5 ± 0.03	0.1	0.3	0.5 ± 0.02	0.3	0.5
20:3n-6	0.1 ± 0.01	0.1	0.1	0.1 ± 0.01	0.1	0.1
20:3n-3	0.1 ± 0.01	0.1	0.1	0.1 ± 0.0	0.1	0.1
Σ trienes	0.7 ± 0.02	0.3	0.5	0.6 ± 0.04	0.5	0.7
18:4n-3	0.9 ± 0.01	0.2	2.1	1.0 ± 0.02	0.2	1.0
20:4n-6	0.5 ± 0.02	0.9	0.5	0.5 ± 0.01	1.0	0.6
20:4n-3	0.5 ± 0.02	0.2	0.3	0.5 ± 0.03	0.2	0.6
Σ poly with 4	1.9 ± 0.04	1.2	2.9	2.0 ± 0.06	1.4	2.2
20:5n-3	23.2 ± 0.3	16.2	14.0	27.3 ± 0.4	16.9	19.7
21:5n-3	0.7 ± 0.05	0.3	1.1	0.8 ± 0.03	0.4	1.3
22:5n-3	0.4 ± 0.01	0.8	0.8	0.5 ± 0.01	0.9	0.8
Σ poly with 5	24.2 ± 0.30	17.1	15.9	28.6 ± 0.5	18.4	21.8
22:6n-3	6.4 ± 0.40	30.0	11.4	8.8 ± 0.2	38.8	10.3

PC = phosphatidylcholine, PE = phosphatidylethanolamine and LPC = lysophosphatidylcholine.

classes (PC, PE and LPC) is presented in Table IV for both male and female stages. As already mentioned for other compounds, the fatty acid profiles of PC and PE were very similar between sexes but differed between categories. PC showed a relative dominance of palmitic acid followed by EPA (> 20%) and to a lesser extent by oleic, DHA, vaccenic, palmitoleic, linoleic, erucic and 22:1n-9 acids. PE displayed a major contribution of DHA followed by palmitic acid, EPA and to a lesser extent vaccenic, oleic, stearic and erucic acids. In contrast to the two previous components, LPC showed major differences between males and females. Saturated acids dominated in males (40.9%) while monoenoic acids dominated in females (44.2%). Palmitic and stearic acid were major contributors in males, while oleic and vaccenic acids showed the highest percentages in females. Among polyunsaturated acids and other minor contributors, DHA, linoleic acid, myristic and branched acids dominated in females, while 18:4n-3 did in males.

Euphausiid fatty acid structure and trophic interactions

PLS discriminant analysis is used to clarify potential trophic interactions. It is based on 42 published profiles of total lipid fatty acids corresponding to the five dominant species of euphausiids in the Southern Ocean. Three significant discriminant functions (DF) could be defined, which accounted for 71% of the total variance and separated four distinct groups of species (group 1: *E. crystallorophias*, group 2: *E. frigida* and *E. triacantha*, group 3: *E. superba* and group 4: *T. macrura*) that tended to have similar fatty acid composition (Fig. 7, top). DF 1 appeared to discriminate Ec (*E. crystallorophias*) while DF 2 separated Ef (*E. frigida*) and Et (*E. triacantha*) from Es (*E superba*). DF 3 singled out the group of Tm (*T. macrura*). Species variation in fatty acids composition is illustrated in the plot of variables and centroids of the four groups (Fig. 7, bottom), which reflected mostly the changes in neutral lipid composition. DF 1 was positively correlated with 18:1n-9, 18:4n-3, 18:3n-3 and group 1, while DF 2 opposed 14:0, 18:3n-3, 20:5n-3 and group 3, to 22:1, 20:1 18:0 and group 2. Projection of the present data for *T. macrura* from the Southern Indian Ocean and *E. vallentini* (Fig. 7, top) within the model system defined by the literature data, showed strong similarities with both group 3 and 4, suggesting that the two species are to a large extent omnivorous during summer.

Discussion

Relationships between size, lipid content and lipid constituents

Allometric relationships between wet weight and body length have been reported for several Antarctic euphausiids but most studies have been devoted to *E. superba* (Kils 1981, Kato *et al.* 1982, Farber-Lorda 1994, Mayzaud *et al.* 1998) and *T. macrura* (Rakusa-Suszczewski & Stepnik 1980, Farber-Lorda 1990, 1994). Usually, allometric exponent values suggested isometric growth (i.e. body weight increases with the cube of length) with values relatively close to 3, with little or no differences according to sex and maturity stage. The data recorded in the present study for *T. macrura* showed a lower exponent value for

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juveniles (2.8 ± 0.2) than for adult (4.4 ± 0.5) indicative of a lower growth in length for juvenile than for adults in summer. The summer growth seemed even lower for *E. vallentini*, which showed allometric exponents ranging from 2.6 (± 0.2) in females to 1.9 (± 0.2) in males.

The relationship between body weight and lipid accumulation is always complex because of the many factors other than size controlling lipid content. In the present work, *T. macrura* showed a significant allometric relationship between total lipids and body weight. In summer, juveniles and to a lesser extent males appeared to show a lower rate of lipid accumulation than females. As anticipated, wax esters accumulated at a faster rate than polar lipids as already noted by Hagen (1988) for individuals collected in the Weddell Sea and around the Antarctic Peninsula. *Euphausia vallentini* also showed a significant allometric relationship between wet weight and lipid but without significant differences between males and females. Contrary to *T. macrura*, the changes in lipid content of *E. vallentini* were mostly related to the increase in polar lipids and to a minor extent in triacylglycerols. Such a leading role for polar lipids has been reported earlier for *Euphausia superba* (Mayzaud *et al.* 1998) for individuals collected in the same region in summer. Phosphatidylcholine was the dominant polar lipid fraction in these dynamics and confirmed the covariation of PC and total lipids in Antarctic euphausiids (Saether *et al.* 1985, Hagen 1988, Hagen *et al.* 1996, Mayzaud 1997, Mayzaud *et al.* 1998). Interaction between PE and size was highly significant ($P = 0.001$) suggesting that the contribution of PE to the changes in total lipids is likely related to the covariation with size and weight.

Changes in lipid content: environmental variability and life cycle

It is now well accepted that lipid content in Antarctic euphausiids varies with season and location in relation with the main features of the life cycle (Falk-Petersen *et al.* 2000). Hence, interpretation of lipid content and lipid structure of a given species across different Antarctic provinces is always difficult when data are limited to a single season. In the case of *T. macrura* the seasonal changes in total lipids and wax esters have been described for three life stages (larvae, immature and adults) by Hagen & Kattner (1998) for the eastern Weddell Sea. Maximum and minimum lipid contents were recorded respectively in fall/summer and late winter. All life stages followed the same seasonal trends without significant differences between males and females. Their summer lipid contents were highly variable and ranged from 33.1 ± 8.6% (dry weight basis) for immature to 39.4 ± 6.7% for adults. Comparison with the present results is of course limited to the summer period, after reproduction has taken place. The lipid content was within the range reported (Hagen 1988, Hagen & Kattner 1998, Falk-Petersen *et al.* 1999) but with lower mean values, i.e. (converted to % dry weight) 7.02% (± 1.5) in males, 12.9% (± 9.0) in juveniles and 22.2% (± 12.1) in females. These differences can be partly attributed to spatial heterogeneity, since females showed a mean maximum value of 31.2% (± 6.8) at station 20 while juveniles showed a mean maximum value of 25.1% (± 11.3) at station 12. The most striking difference is the extremely low lipid content observed in the present study for males collected in very small numbers at all four stations. High mortality and lipid depletion in sexually active males has been reported by Virtue *et al.* (1996) for *E. superba* but direct influence of mating activity in *T. macrura* is unlikely since it took place in late winter (Stepnik 1982). Further data are needed to evaluate whether our observations corresponded to an occasional event or a repetitive site-specific feature. Similar comparison for *E. vallentini* showed a fair agreement with the value reported by Attwood & Hearshaw (1992) for the Prince Edward Islands area in late summer–early autumn.

The presence of high levels of wax esters in *T. macrura* has been reported earlier by Reinhardt & Van Vleet (1986) and confirmed by Hagen (1988). Accumulation of wax esters in zooplankton has been related to high seasonality in light regime and food supply (phytoplankton) which is maximum at high latitudes (Lee & Hirota 1973, Sargent & Henderson 1986). As discussed by Båmstedt (1986), such massive storage of metabolic energy seems to be an adaptive response of high latitude zooplankton, which allows initiation of breeding independently of primary production. As a direct consequence, it has been further suggested that accumulation of wax esters concerned mostly herbivorous species and that carnivores or detritivores did not store quantities of wax ester as large as those in herbivores (Sargent & Henderson 1986). The relationship between high concentration of wax esters and high reproductive requirements during periods of low food supply also applies to Antarctic euphausiids. Indeed, species with extensive wax esters storage, such as *Euphausia crystallorophias* and *T. macrura*, are known to initiate sexual maturation in winter or fall before spring biological production (Stepnik 1982, Nordhausen 1994), while *Euphausia superba* and *Euphausia vallentini*, which accumulate only triacylglycerols during summer, rely on spring and summer primary production for sexual maturation and spawning (Makarov 1979). Summer content of wax esters (% of total lipids) have been shown to range from 54.9 ± 11.9% for immature *T. macrura* to 62.4 ± 4.8% in adults collected in the Weddell Sea (Hagen & Kattner 1998) and from 45 to 50% for unstaged individuals collected in the Lazarev Sea (Falk-Petersen *et al.* 1999). The present study suggested a lower level of wax ester accumulation, but with a high degree of spatial variability. The maximum percentages recorded were closer to the range reported by previous authors with 53.0 ± 0.6% for

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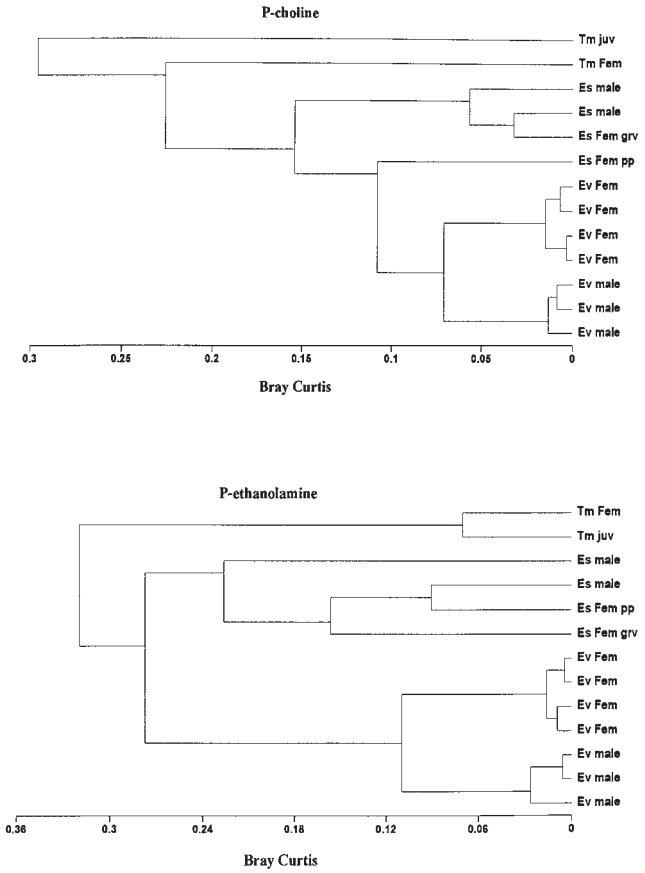

Fig. 8. Classification by clustering of *E. vallentini*, *T. macrura* and *E. superba* based on the fatty acid composition of two phospholipid constituents: phosphatidylcholine (top) and phosphatidylethanolamine (bottom). Data for *E. vallentini*, *T. macrura* were from this study and data for *E. superba* from Mayzaud (1997).

females (station 20) and 30.8 ± 3.6% for juveniles (station 12). There is no reason to assume that the life cycle described by Hagen & Kattner (1998) does not apply to other areas of the Southern Ocean and, local differences in trophic interactions and hydrodynamic regime were probably responsible for the observed variability. Spatial heterogeneity may also explain the differences in wax ester fatty acid and fatty alcohol composition observed between the various studies. The higher percentages, in juveniles than in female, of 18:1n-7 alcohol as well as the lower content of EPA found in the present work contrasts with the opposite pattern reported by Hagen & Kattner (1998).

Position of T. macrura *and* E. vallentini *in the food web*

In terms of trophic interactions, euphausiids encompass a large spectrum of feeding habits. *E. crystallorophias* and *E. superba* are considered primarily herbivorous but are known to be able to resort to carnivory or omnivory during non bloom periods (Kittel & Ligowski 1980, Hopkins 1987, Atkinson & Snyder 1997). *Thysanoessa macrura* has been shown to be more omnivorous (Mayzaud *et al.* 1985, Hopkins & Torres 1989), while other more northern species such as *E. triacantha* and *E. frigida* are considered carnivorous (Phleger *et al.* 2002). Little is known of the large-scale variability of these feeding patterns and the actual trophic status of *E. vallentini* is virtually unknown. Fatty acid composition of total or neutral lipids have been often used in the interpretation of major trophic interactions. Different ratios and relationships have been proposed to illustrate the dominant feeding regime. Cripps & Atkinson (2000) proposed to use the PUFA/SFA ratio of total lipids as an index of carnivory while others (Phleger *et al.* 2002) used EPA/DHA or 18:1n-7/18:1n-9. Most of these ratios are useful but showed limited predictive capability for large-scale comparisons. To aid in the interpretation of the feeding pattern of Antarctic euphausiids, we used a multivariate approach, i.e. PLS discriminant analysis. The system of discriminant functions observed (Fig. 7) can be interpreted in terms of dominant feeding behaviour. Discriminant function 1 (DF 1) suggested a gradient from mostly herbivory (*E. crystallorophias*) to omnivory (*E. superba* and to a minor extent *T. macrura*) while DF 2 suggested a gradient between omnivory (*E. superba*) to carnivory (*E. triacantha* and to a minor extent *E. frigida*). The high variability observed in some of the grouping is likely to, in part, reflect the local changes in feeding patterns. Hence, Et1, Et2 and Ef1 (from Phleger *et al.* 1998) are mostly carnivorous while Et3 (from Cripps & Atkinson 2000), Et4, Et5 and Ef2 (from Phleger *et al.* 2002) were far more omnivorous. The same line of interpretation indicated that *E. vallentini* has a similar level of omnivory to *E. superba* while the *T. macrura* sampled in the Indian sector of the Southern Ocean appeared more omnivorous than their Weddell Sea or Ross Sea counterparts. The high degree of variability may also be explained by the use of total lipid fatty acid profiles, since essentially the neutral lipids are directly linked to the structure of the lipid ingested and the trophic relationships are somewhat confused by the phospholipid fatty acid constituents mostly related to internal metabolic processes and genetic control.

Polar lipids: a role in reproductive processes

Despite their leading roles in all aspects of the metabolism, the changes in structure of euphausiid polar lipids have received limited attention. In fact data on the fatty acid structure of PC and PE is limited to *E. superba*. Earlier reports (Van der Veen *et al.* 1971, Clarke 1980) suggested higher levels of HUFA (20:5+22:6) in PE than in PC and lower percentages of palmitic acid (16:0). Later data by Mayzaud (1997) and Mayzaud *et al.* (2000) suggested a different pattern with higher percentages of PUFA, 18:4n-3, 18:3n-3 in PC than in PE for gravid females and males but

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not for spent females, which showed a severe decrease in the level of 22:6n-3 for PC. Conversely, high percentages of 18:1n-7 appeared typical of PE for all stages of *E. superba*. A comparison of the PC and PE fatty acid structures of the data reported in the present study and those for *E. superba* (Mayzaud 1997) was made using a cluster analysis based on the Bray and Curtis distance index (Fig. 8). For both polar lipid classes the fatty acid profiles were well classified according to species with *T. macrura* being most distinct. One interesting exception was noticeable for the PC fraction with *E. vallentini* displaying strong similarities with *E. superba* spent females because of the depletion of 22:6n-3 (< 10% of fatty acid pool) compared to 20:5n-3. The key role of 22:6 in the reproductive process has been suggested by Virtue *et al.* (2000) who showed a higher content in reproducing females of the northern krill *Meganyctiphanes norvegica*. If the same applies for Antarctic euphausiids, the samples of *E. vallentini* females analysed were essentially post spawn at the time of capture, as suggested by the annual cycle study carried out in a similar ecosystem by Ridoux (1988).

In conclusion, although the two species of euphausiids studied showed different lipid patterns both in terms of the nature of lipid accumulated and lipid metabolism, they both confirmed the strong link between lipid dynamics and reproduction strategies. *Euphausia vallentini* showed strong similitude with *E. superba*, which also accumulates triglycerides and reproduces during the favourable summer feeding conditions. The link between reproduction and polar lipids was illustrated for both species with the reduction of DHA in the PC fraction of *E. vallentini* and the depletion of PE in *T. macrura*. The involvement of phospholipids in ovary development and egg production of zooplankters has probably been often overlooked and needs further investigation.

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